

Individual histories and selection in heterogeneous populations

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Edited* by David R. Nelson, Harvard University, Cambridge, MA, and approved May 28, 2010 (received for review October 29, 2009)

The strength of selection in populations has traditionally been inferred by measuring changes in bulk population parameters, such as mean reproductive rates. Untangling the effect of selection from other factors, such as specific responses to environmental fluctuations, poses a significant problem both in microbiology and in other fields, including cancer biology and immunology, where selection occurs within phenotypically heterogeneous populations of cells. Using “individual histories”—temporal sequences of all reproduction events and phenotypic changes of individuals and their ancestors—we present an alternative approach to quantifying selection in diverse experimental settings. Selection is viewed as a process that acts on histories, and a measure of selection that employs the distribution of histories is introduced. We apply this measure to phenotypically structured populations in fluctuating environments across different evolutionary regimes. Additionally, we show that reproduction events alone, recorded in the population’s tree of cell divisions, may be sufficient to accurately measure selection. The measure is thus applicable in a wide range of biological systems, from microorganisms—including species for which genetic tools do not yet exist—to cellular populations, such as tumors and stem cells, where detailed temporal data are becoming available.

phenotypic diversity | selection strength | statistical mechanics | stochastic switching | fundamental theorem of natural selection

Measuring the strength of selection in populations is fundamental to any quantitative description of evolution. In laboratory experiments, populations of microorganisms can be propagated for many generations, in constant or fluctuating conditions, and adaptation of growth rates and other characteristics can be measured (1). Adaptation of the population as a whole may arise from individual responses, such as sensor-mediated changes in gene expression activating specific pathways that are beneficial in certain conditions. Likewise, it can result from selection acting on existing, heritable phenotypic and/or genotypic differences between individuals [e.g., as in antibiotic persistence (2), bacterial sporulation and competence (3), and phase variation (4)]. In reality, both individual responses and population-level selection occur concurrently, and the adaptation of the population results from the mixture of these two forces. It should be highly informative therefore to characterize the *effective* strength of selection in experimental evolution. Such measurement would identify specific environmental conditions that require adaptation via selection, as well as those for which an organism already possesses suitable genetic pathways of response. It could, in principle, be predictive as well of what an organism can easily adapt to via selection, and what might be more difficult.

In population genetics studies, the existence of selection and its strength are indirectly inferred from analysis of existing genetic variation in populations (5). In experimental evolution, where one hopes to directly observe selection in action, selection measurements have been based on changes in bulk population growth rate and on the variance of reproductive rates (1). The insight of Fisher (6) was to partition the change in the population growth

rate into two terms, the first due to changes in allele frequencies, and the second due to changes in environmental conditions (7–10). The first term was defined as the measure of selection and was proven to be equal to the population’s variance of fitness. While mathematically valid, and intuitively appealing, the theorem is difficult to apply directly to experiments because it neglects the effect of mutations, as well as other aspects of population structure such as phenotypic heterogeneity, specific responses, and environmental fluctuations, all of which are relevant on time scales of laboratory experiments.

Recently, detailed temporal information about individuals in microbial populations has become available (3, 11–15). Using gene-specific fluorescent reporters, video microscopy, and automated image analysis, one can follow each “history,” i.e., temporal sequences of all reproduction events and all phenotypic changes of a given individual and of its ancestors (16). Cell lineage data are also becoming available in other systems, including hematopoietic stem cells (17) and carcinoma cell lines (18). Such detailed data should allow one to proceed beyond the classical measures of selection, both in microbiology and in other fields such as cancer biology where selection occurs within phenotypically heterogeneous populations of cells (19, 20). We introduce here a theoretical framework that takes full advantage of individual-level temporal data that is typical of recent experiments, while simultaneously maintaining the intuitive aspects of Fisher’s and Price’s formulations of population evolution (6, 21). Key to our approach is the shift of focus from individual organisms to individuals’ histories.

What needs to be measured regarding selection? Selective differences are certainly measurable, when sufficiently large, so this poses no fundamental problem. How such differences propagate to the population level, however, can depend strongly on mutation rates, phenotypic heterogeneity, environmental fluctuations, population sizes, and demography. Finding the key measurements of this process is thus at the heart of what selection means as a biological concept. To avoid a potentially subjective resolution of this problem, we introduce here a thought experiment that provides a conceptual basis for measuring selection, which is inspired by similar problems in the physical sciences. To gauge the importance of thermal fluctuations, for example, for the behavior of a physical system, such as a solution, or a crystal, the natural approach is to change temperature and measure how the system responds. Similarly, to gauge the importance of selective differences for the behavior of a population, it seems natural to change the relative magnitude of those differences and measure how the

Author contributions: S.L. and E.K. designed research; S.L. and E.K. performed research; and S.L. and E.K. wrote the paper.

The authors declare no conflict of interest.

*This Direct Submission article had a prearranged editor.

Freely available online through the PNAS open access option.

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This article contains supporting information online at www.pnas.org/lookup/suppl/doi:10.1073/pnas.0912538107/-DCSupplemental.

Analogous to the population mean fitness, in the history formulation we define the mean fitness of histories, \bar{H} :

$$\bar{H} = \sum_{\sigma} H_{\sigma} x_{\sigma} \quad (\text{historical mean fitness}).$$

Selection and Histories. To decouple selection from other effects, the history formulation allows us to change β and measure the response of any quantity. We compute how a change of β affects the frequency of histories,

$$\frac{\partial x_{\sigma}}{\partial \beta} = \frac{P_{\sigma} H_{\sigma} e^{\beta H_{\sigma}}}{N} - \frac{P_{\sigma} e^{\beta H_{\sigma}}}{N^2} \frac{\partial N}{\partial \beta} = (H_{\sigma} - \bar{H}) x_{\sigma}, \quad [1]$$

and how the change of β affects the mean historical fitness,

$$\frac{\partial \bar{H}}{\partial \beta} = \sum_{\sigma} H_{\sigma} \frac{\partial x_{\sigma}}{\partial \beta} = \text{Var}(H_{\sigma}) \quad (\text{historical fitness relation}).$$

Returning to the thought experiment, the left-hand side above corresponds to the result of comparing experiment B to experiment A. Evaluating the result at $\beta = 1$ (i.e., a tiny perturbation of fitness), the right-hand side is then precisely the historical fitness variance in experiment A. Therefore we can assert that the result of the thought experiment is, in fact, measurable without performing experiment B.

The use of the derivative with respect to β to measure the effective strength of selection is natural, as we have argued based on an analogy with physical systems (see *Discussion*). Derivatives of quantities other than \bar{H} , however, may, in principle, be useful as well. For example, the population mean fitness \bar{f} is another natural candidate, which can be written as $\bar{f} = \sum_{\sigma} f_{\sigma(t)} x_{\sigma}$, whose derivative is found using Eq. 1 to be $\partial \bar{f} / \partial \beta = \text{Cov}(f_{\sigma(t)}, H_{\sigma})$. Here, we will use the historical fitness variance per unit time,

$$\mathcal{M}_s \equiv \left. \frac{1}{t} \text{Var}(H_{\sigma}) \right|_{\beta=1} \quad (\text{measure of selection}),$$

as the measure of selection. We make this choice due to the formal similarities between \mathcal{M}_s and Fisher's result, as well as the direct applicability of \mathcal{M}_s to experiments, which will be shown below.

We can interpret the historical fitness relation in terms of the behavior of histories. Eq. 1 shows that upon a change of β , frequencies of histories change as though the histories themselves were the fundamental replicating entities. Histories with larger fitness will benefit from a change in historical conditions exponentially more than histories with smaller fitness. If histories in the population have similar values of H_{σ} , the change in historical conditions benefits them equally, and the mean historical fitness, \bar{H} , changes minimally. Conversely, if the distribution of H_{σ} is broad, the change in historical conditions benefits those histories with high H_{σ} disproportionately, and the change in the mean historical fitness is large. The intuition behind Fisher's theorem is therefore applicable to histories as replicating entities.

Results

To investigate the behavior of histories across different evolutionary regimes, we now specialize the general model. Individuals will be assumed to have two phenotypic states, each adapted to two different environments. Environmental changes occur here periodically, with period τ , and individuals can switch phenotype in a way that is either *stochastic* (switching randomly between the phenotypes) or *responsive* (switching specifically to the adapted phenotype, i.e., sensing). Adapted individuals reproduce with rate f_a , and nonadapted individuals at a lower rate f_{na} . Rates

of stochastic and responsive switching are given by s and s_r , respectively. The model allows for organisms that employ a mixture of stochastic and responsive switching, organisms that sense and respond but might make errors, as well as pure stochastic ($s_r = 0$) and pure responsive ($s = 0$) organisms.

We simulate these stochastic and responsive models, keeping track of individual histories, for both normal ($\beta = 1$) and improved ($\beta = 1.1$) historical conditions. Upon a change of historical conditions, the change in \bar{H} observed in simulation is used to estimate \mathcal{M}_s , as shown in Fig. 2A. Even with very few histories depicted in Fig. 2, it is apparent that the larger the historical fitness variance in normal conditions, the larger the increase in mean fitness upon improved conditions (see also Fig. S1). To verify the relation quantitatively, $\text{Var}(H_{\sigma})$ must be estimated accurately (as in Fig. 3 below), which requires hundreds of independent histories (see *Discussion*).

The value of \mathcal{M}_s can be computed exactly (see *Methods*), for any instance of the given model, over the entire range of fluctuation periods τ (Fig. 3). The strength of selection measured by \mathcal{M}_s depends on both the nature of individual behavior and the period of environmental fluctuations. For stochastic switching (Fig. 3A), a pronounced peak in \mathcal{M}_s (note the logarithmic scale) is present at values of τ significantly larger than the generation time. For responsive switching (Fig. 3B), the \mathcal{M}_s curve is shifted downward relative to the pure stochastic case, and its peak becomes less pronounced. The existence and meaning of these peaks can be understood qualitatively (see *Discussion*), by considering the relevant population dynamics. Likewise, asymptotic behaviors of these curves can be derived in the limits of fast ($\tau \ll 1$) and slow ($\tau \gg 1$) environmental fluctuations (see *SI Text*). We note that \mathcal{M}_s clearly distinguishes between stochastic and responsive switching via the location and magnitude of the peak with respect to the dashed line $1/\tau$. Also in agreement with our expectation, increasingly slow responsive switches ($s_r \ll 1$) behave increasingly like pure stochastic switches, as their peak crosses this line. Fig. S2 presents additional plots for cases of fast stochastic switching, and switching with asymmetric fitness values.

The definition of \mathcal{M}_s based on detailed phenotypic histories seems to imply that measurement of this quantity directly from experimental populations (or in this work, directly from simulated ones) would require information about the internal phenotypic states of cells. Surprisingly, however, we find that \mathcal{M}_s can in some cases be inferred accurately using only the distribution of cell divisions over individual histories. Indeed, visual inspection of Fig. 2B shows that for slow stochastic switching, for which the effective selection strength \mathcal{M}_s is large, the pattern of cell divisions across the histories is strikingly nonuniform, with patches of cell division separated by empty intervals (in contrast to responsive switching).

This observation is explained by noticing that the number of cell divisions observed in a history σ is a random variable D_{σ} whose statistical properties depend on the cell's historical fitness H_{σ} . Variance in D_{σ} between histories results from two sources: (i) variance of the historical fitness of different cells, i.e., $\text{Var}(H_{\sigma})$, and (ii) variance of the cell division process itself. The latter is given by \bar{D}_{σ} , for the case in which the times between cell divisions are exponentially distributed random variables. This results in the following measurement formula (see *SI Text*): $\mathcal{M}_s = (1/t)[\text{Var}(D_{\sigma}) - \bar{D}_{\sigma}]$.

By performing independent replicate experiments, in which all cell divisions are recorded, \mathcal{M}_s may thus be measured using the difference between the variance and the average number of cell divisions over independent histories, with a large difference resulting in the patchiness observed in Fig 2B. Fig. 3 depicts the results of simulations, in which we measured \mathcal{M}_s from cell division statistics (filled circles), showing an excellent agreement with values expected from exact calculations (solid curves). With a modification, the same approach may be used to measure \mathcal{M}_s also

tion between evolutionary dynamics and the theory of disordered systems, such as spin glasses (29). Spatial fluctuations in growing populations were considered via analogy with convection by Nelson and Shnerb (30). Recently, several groups have described evolution of allele frequencies in populations using physical analogies, both from equilibrium and nonequilibrium points of view (32, 33).

To characterize the interplay between mutation and selection, Baake and co-workers introduced the idea of the ancestral distribution of types along histories (23) and later, using the theory of large deviations, considered competitions between lines of descent having different ancestral distributions in a constant environment (reviewed in ref. 34). As an alternative to large deviations, we previously presented a mapping between population dynamics and the theory of heteropolymers, which maps polymer conformations to histories, and predicted a population phase transition that occurs as environmental durations τ change (31). More recently, Mustonen and Laessig considered temporal trajectories of the frequency distribution, $n_i(t)$, as realizations of nonequilibrium dynamics in finite populations, and introduced a measure of population adaptation called the fitness flux, Φ , which considers the rate of frequency changes multiplied by their fitness value (33). Because this approach is based on population trajectories, rather than individual histories, intriguing possibilities exist of bridging these two viewpoints.

The most important departure between the history formulation described here and previous studies is the introduction of the historical fitness relation as a tool to measure selection. Remarkably, at least in the simple cases we have examined, enough information exists in the distribution of cell divisions along individual histories for direct measurement of \mathcal{M}_s , without knowledge of individuals' phenotypic states and fitness values. This opens possibilities of precisely measuring the effect of selection in controlled experiments involving populations of microbes, other organisms, or individual cells.

Methods

Analytical Calculation of \mathcal{M}_s . The distribution of H_σ (evaluated at $\beta = 1$) has the following cumulant generating function: $K(\mu) = \log \sum_\sigma x_\sigma e^{\mu H_\sigma} =$

$\log N(t, 1 + \mu) - \log N(t, 1)$. If κ_n is the n th cumulant of the historical fitness distribution, then we have

$$\kappa_n = \left. \frac{\partial^n K(\mu)}{\partial \mu^n} \right|_{\mu=0} = t \left. \frac{\partial^n \tilde{\Lambda}}{\partial \beta^n} \right|_{\beta=1}; \quad [3]$$

i.e., $\tilde{\Lambda}$ is the cumulant generating function of historical fitness. To evaluate \mathcal{M}_s , we find $\tilde{\Lambda}$, take two β derivatives, and evaluate at $\beta = 1$. For the periodic case of two environments considered in the text, we define the matrices A_1 and A_2 by $(A_k)_{ij} \equiv \beta f_i^k \delta_{ij} + s_{ij}^k$ (where δ_{ij} is the Kronecker delta symbol), and find $\tilde{\Lambda} = (1/2\tau) \log \lambda_1 [e^{A_1} e^{A_2}]$, where λ_1 denotes the maximum eigenvalue of the matrix, as in ref. 35. For the case of two-by-two matrices, this is computable exactly, as is $\mathcal{M}_s = \partial^2 \tilde{\Lambda} / \partial \beta^2$ (evaluated at $\beta = 1$), shown in Fig. 3.

Simulation Method and Bounded Populations. Stochastic simulations of population dynamics were performed, in which all cell divisions were recorded, and all individuals' histories were stored. We used the two-state model described in *Results*, simulating the continuous-time stochastic processes for a small population as in ref. 35. To avoid double counting, each division is assigned arbitrarily to one and only one lineage (see *SI Text*). Each time the population size reached 1,000 cells, only 100 randomly chosen cells were allowed to continue. Simulation length was 3,000 time units, and data from $n_{\text{runs}} = 1,000$ simulations were used to obtain each data point in Fig. 3C. The number of histories present in a bounded population comprises a tiny fraction of the total number of possible histories over any time interval. These histories are correlated due to common descent. To obtain independent histories for averaging, from each of the n_{runs} simulations a single individual history and a time window of size t_w were chosen randomly. The measurement formula for \mathcal{M}_s was applied to the n_{runs} windows (with $t = t_w$), to obtain one estimate of \mathcal{M}_s . The entire random windowing process was repeated 100 times (i.e., resampling from the same simulation data), and the estimates of \mathcal{M}_s were averaged, to obtain the final estimate. The results shown in Fig. 3 used a window size of $t_w = 100$ time units.

ACKNOWLEDGMENTS. We thank Alexander Grosberg, David Huse, Bruce Levin, Henri Orland, Richard Losick, Seppe Kuehn, Olivier Rivoire, Amoolya Singh, Madan Babu, Doeke Hekstra, and James Bull for invaluable conversations regarding this work. E.K. thanks the Burroughs-Wellcome Fund Career Award at the Scientific Interface for financial support.

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